(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) EP 0 496 818 B1

(12) EUROPEAN PATENT SPECIFICATION

(45) Date of publication and mention of the grant of the patent:
23.06.1999 Bulletin 1999/25

(21) Application number: **90916618.3**

(22) Date of filing: **19.10.1990**

(51) Int Cl.[6]: **C07K 16/28**, A61K 39/395, C12P 21/08

(86) International application number:
PCT/US90/06030

(87) International publication number:
WO 91/05805 (02.05.1991 Gazette 1991/10)

(54) MONOCLONAL ANTIBODY SPECIFIC FOR IgA RECEPTOR

IGA-REZEPTORSPEZIFISCHE MONOKLONALE ANTIKÖRPER

ANTICORPS MONOCLONAL SPECIFIQUE POUR LEUR RECEPTEUR IgA

(84) Designated Contracting States:
AT BE CH DE DK ES FR GB IT LI LU NL SE

(30) Priority: **20.10.1989 US 424883**

(43) Date of publication of application:
05.08.1992 Bulletin 1992/32

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Printed by Jouve, 75001 PARIS (FR)

Description

Background

[0001] Receptors for the Fc portion of immunoglobulin are important in triggering many of the protective functions of monocytes, macrophages and polymorphonuclear cells. While receptors for IgG on these cells have been extensively investigated, it is becoming evident that receptors for IgA are also capable of promoting effector functions of these cells and that IgE may stimulate some activities of monocytes. While soluble IgA binds IgA receptor with poor avidity, polymerized IgA has been demonstrated to trigger functions such as superoxide generation and phagocytosis.

Summary of the Invention

[0002] This invention pertains to monoclonal antibody which specifically binds to Fc receptor for IgA (Fc-alpha receptor) on an effector cells such as a monocytes, polymorphonuclear cells and macrophages and which can trigger Fc-alpha-receptor-mediated effector function. The antibody (or fragment thereof) can be linked (chemically or genetically) to an antibody (or fragment thereof) specific for a target antigen to form a bispecific antibody or heteroantibody. These bispecific molecules can be used direct effector cells to cell bearing the target antigen, resulting in cytolysis of the cell.

Detailed Description of the Invention

[0003] The antibody of this invention binds the Fc-alpha receptor (FcRI) for human IgA. The monoclonal anti-Fc-alpha receptor antibody of this invention can be produced by conventional monoclonal antibody methodology e.g., the standard somatic cell hybridization technique of Kohler and Milstein, Nature 256: 495 (1975). Although somatic cell hybridization procedures are preferred, in principle, other techniques for producing monoclonal antibody can be employed e.g., viral or oncogenic transformation of B lymphocytes.

[0004] Human cells bearing Fc-alpha receptor can be used to immunize an animal for production of monoclonal antibody. Alternatively, the receptor for immunization of an animal can be prepared from lysates of human cells which express the receptor, e.g., a human monocytic cell. In another mode, a partially purified preparation of the receptor can be made by lysing receptor-bearing cells and then purifying the receptor by immunoadsorbant chromatography. Cells can be lysed in a buffer containing a detergent such as NP40. The immunoadsorbent can be prepared by attaching human IgA to a water-insoluble material such as an activated Sepharose™ resin. The Sepharose resin with attached human IgA is poured into a column. The cell lysate is passed through the column under conditions which permit adsorbtion of the cellular Fc receptor protein by the IgA coupled to the resin. The adsorbed Fc receptor protein can be eluted with a mildly acidic elution buffer. The purified receptor can then be used for immunization of an animal to produce anti-receptor monoclonal antibody.

[0005] The preferred animal system for preparing hybridomas is the murine system. Hybridoma production in the mouse is a very well-established procedure. Immunization protocols and techniques for isolation of immunized splenocytes for fusion are well known in the art. Fusion partners (e.g., murine myeloma cells) and fusion procedures are also well-known.

[0006] Employing the methodology described, a monoclonal antibody (mAb) My 43 of the IgM class which binds specifically to monocyte and polymorphonuclear cell IgA receptors, based on its ability to block IgA mediated rosettes and phagocytosis. This antibody recognizes a surface molecule which triggers function since monocytes and PMNs secrete superoxide when treated with this antibody. Additional technical information on My 43 is reported in Shen, L., Immunology 68:491-496 (1989) and Shen, et al., J. Immunol. 143(12):4117-4122 (1989).

[0007] The above referenced articles describe the following method of producing the antibodies of the present invention:

[0008] The antibodies of this invention can be used to target effector cells bearing Fc-alpha receptor. To target effector cells, bifunctional antibodies or heteroantibodies are employed. These antibodies have dual antigen binding specificity - one specificity for the Fc-alpha receptor and one specificity for an epitope of the target cell. The Fc receptor specificity mediates linkage to the effector cell through a known cytotoxic trigger molecule. The target cell specificity provides for recognition and binding to the target cell.

[0009] Bifunctional antibodies are single, divalent antibodies which have two different antigen binding sites. Bifunctional antibodies for targeting have one binding site for Fc receptor and one binding site for a target cell epitope.

[0010] Heteroantibodies are two or more antibodies or antibody binding fragments (Fab) linked together, each antibody or fragment having a different specificity. Heteroantibodies for targeting comprise an antibody or antigen binding fragment specific for Fc receptor for IgA, coupled to an antibody or antigen binding fragment thereof specific for a target cell epitope.

Myeloid Cell Lines

[0011] The monocyte-like cell line U-937 was maintained in continuous culture in RPMI-1640 (supplemented with 10% FCS). Differentiation was induced by addition of 300 IU/ml of IFN-γ (kindly supplied by Genentech, South San Francisco, CA), and cells were assayed 4 days posttreatment. Before subsequent experimentation, the cells were washed three times in PBS. HL-60 cells were maintained in serum-free Iscove's modified Dulbecco's medium (13). To promote IgA receptor expression cultures were supplemented with 10^{-7} M I-25 dihydroxy-vitamin D_3 (a generous gift of Hoffman LaRoche, Nutley, NJ) and kept at 1 to 2 x 10^6 cells/ml for 7 days.

Monocytes

[0012] Mononuclear cells enriched for monocytes were obtained by cytopheresis of peripheral blood from normal volunteers. These cells, which usually contained >50% monocytes, were further purified by centrifugation over Ficoll Hypaque (Pharmacia Fine Chemicals, Piscataway, NJ). The mononuclear cell layer was washed five times in RPMI 1640 medium and resuspended in 10% FCS at 5 x 10^7 cells/ml in a 50 ml polypropylene tube. The tube was rotated for 90 min at 4°C, causing the monocytes to clump, and was then placed upright in an ice bath for 30 min. allowing the clumped cells to settle as a pellet. Sedimented cells were washed three times in RPMI 1640 medium. and usually contained greater than 90% monocytes, the remainder being lymphocytes (14).

Human Ig

[0013] Human dimer IgAl paraproteins (Sa IgA and Ca IgA were purified from myeloma sera by gel chromatography on Sepharose CL-6B (Pharmacia Fine Chemicals). The peak corresponding to dimer IgA was cleared of contaminating IgG on protein A-Sepharose CL-6B; these preparations contained less than 0.5 µg IgG/mg IgA by ELISA (Southern Biotechnology Associates, Birmingham, AL). Human IgGl was purified from the plasma of a myeloma patient by ion-exchange chromatography on an Affi-Blue Gel column (Bio-Rad, Richmond, BALB/c mice were immunized with 1 X 10^7 monocytes at 10-day intervals. Five days after the second immunization they were killed and spleen cells hybridized with NS-1 nonsecreting myeloma cells as previously described (15). Resulting hybridomas were tested for production of antimonocyte mAb by indirect immunofluorescence. Antimonocyte mAb were tested for blocking of FITC-IgA binding to monocytes as described below (see Immunoflurescence). The hybridoma producing the only mAb that blocked IgA binding. My 43, was subcloned twice by limiting dilution. My 43 was determined to be of the IgM isotype by ELISA (Southern Biotechnology Associates). Ascites were produced by injecting 5 x 10^6 cells i.p. into pristane primed BALB/c mice. IgM was purified from ascites by HPLC gel filtration by using a TSK 400 column (Bio Rad) followed by passage through protein A-Sepharose 4b (Pharmacia).

Hybridoma Supernatants

[0014] Hybridoma cell lines were maintained in Dulbecco's medium containing 10% FCS. Supernatants were collected twice a week and pooled into batches that were tested for antibody presence by immunofluorescence. To determine actual antibody content, supernatants were concentrated tenfold using Centricell (Polysciences Inc., Warrington, PA) and compared to IgG or IgM standards in Diffugen radial immunodiffusion plates (Tago, Burlingame, CA). Supernatants containing 10 to 20 µg/ml mAb were used in experiments.

[0015] Bifunctional antibodies can be produced by chemical techniques (see e.g., D. M. Kranz et al., Proc. Natl. Acad. Sci. USA 78,5807 (1981)) by "polydoma" techniques (See U.S. Patent 4,474,893, to Reading) or by recombinant DNA techniques. Heteroantibodies can be prepared by conjugating Fc receptor antibody with antibody specific for an epitope of a target cell. A variety of coupling or crosslinking agents can be used to conjugate the antibodies. Examples are protein A, carboiimide, and N-succinimidyl-3-(2-pyridyldithio) propionate (SPDP). SPDP is the preferred agent: procedures for crosslinking antibodies with this agent are known in the art. See e.g., Karpovsky et al., (1984) J. Exp. Med. 160:1686; Liu, M.A. et al., (1985) Proc. Natl. Acad. Sci USA 82:8646.

[0016] Employing the SPDP agent, bi-Specific antibodies of the monoclonal antibody My 43 and Fab anti-erythrocyte antibodies were prepared and shown to promote phagocytosis by monocytes (whereas bi-specific antibodies of anti-RBC x-anti-beta$_2$ microglobulin did not). In comparative studies on phagocytosis, an average of 52% of monocytes ingested IgKG coated red cells and 32% ingested cells coated with My 43 bi-specific antibodies.

[0017] Target cells are cells whose elimination would be beneficial to the host. One important type of cell is a tumor cell. Effector cells can be targeted with bifunctional or heteroantibody having specificity for FcRI and specificity for a tumor associated or tumor specific antigen.

[0018] Antibodies with a desired tumor specificity for production of bifunctional antibody or heteroantibody can be produced or can be selected from available sources. Monoclonal antibodies against tumor-associated antigens can

be made by the methods of Koprowski et al., U.S. Patent 4,172,124. Many suitable anti-cancer antibodies are presently available.

[0019] Specific anti-tumor antibodies would include, but not be limited to:

Antibody	Specificity
AML-2-23, PM-81, PMN-6, PMN-19	Myeloid Leukemia
SCCL-1, SCCL-175	Small Cell Carcinoma of the Lung
OC1-25, OVCT-3	Ovarian Carcinoma
COL-1, COL-2, COL-3, ... COL-13	Colon Carcinoma

[0020] In addition to tumor cells, the effector cell can be targeted against auto-antibody producing lymphocyte for treatment of autoimmune disease or an IgE-producing lymphocyte for treatment of allergy. The target can also be microorganism (bacterium or virus) or a soluble antigen (such as rheumatoid factor or other auto-antibodies).

[0021] Effector cells for targeting are human leukocytes, preferably macrophages. Other cells would include monocytes and other IgA-receptor bearing cells. If desired, effector cells for targeting can be obtained from the host to be treated.

[0022] The targeted effector cells can be administered as a suspension of cells in a physiologically acceptable solution. The number of cells administered can be in the order of 10^8-10^9 but will vary depending on the therapeutic purpose. In general, the amount will be sufficient to obtain localization at the target cell and to effect target cell killing by antibody dependent mediated cytolysis (ADCC). Routes of administration can also vary. In tumor therapy, for instance, depending upon the localization of a tumor, the targeted effector cells could be administered intravenously, or directly into tumor sites; as for example, directly into the peritoneal cavity in the case of ovarian carcinoma.

[0023] Therapy with targeted effector cells can be performed in conjunction with other techniques for removal of targeted cells. For example, anti-tumor therapy with bifunctional antibodies and/or effector cells armed with bifunctional (hetero)antibody can be used in conjunction with surgery, chemotherapy or radiotherapy. Additionally, combination immunotherapy may be used to direct two distinct cytotoxic effector populations toward tumor cell rejection. For example, anti-tumor antibodies linked to anti-Fc-gammaRI or anti-T3 (will trigger cytolytic T lymphocytes to lyse tumor cells) may be used in conjunction with IgA-receptor specific heteroantibodies. Protocols based on these concepts may be especially effective in removing residual tumor cells in patients induced into remission by chemotherapy and irradiation.

[0024] The anti-Fc-alpha receptor antibody of this invention has additional utility in therapy and diagnosis. The Fc receptor antibody itself can be a targeting antibody (i.e., to target for cells bearing Fc-alpha receptor). For example, the antibody can be used to target lipid vesicles containing anticancer drugs for treatment of certain hematological cancers (e.g. acute myeloid leukemia), or to target lipid vesicles containing factors (such as gamma-IFN) which activate monocytes. The antibody, if of the appropriate murine IgG subclass (e.g., IgG2a), can be used directly in vivo to eliminate Fc-alpha-receptor-bearing cells (e.g., myeloid leukemia cells) via natural complement or ADCC mechanisms.

Equivalents

[0025] Those skilled in the art will recognize, or be able to ascertain using no more than routine experimentation, many equivalents to the specific embodiments of the invention described herein. Such equivalents are intended to be encompassed by the following claims.

Claims

Claims for the following Contracting States : AT, BE, CH, LI, DE, DK, FR, GB, IT, LU, NL, SE

1. A monoclonal antibody, or active fragment thereof, specific for human Fc receptor for IgA which antibody can trigger Fc-receptor-mediated effector cell activity.

2. A method of producing the monoclonal antibody of claim 1 or an active fragment thereof, comprising the sequential steps of:

    a. immunizing an animal, e.g. a mouse with human cells bearing Fc-alpha receptor;

b. preparing hybridomas of anti monocyte monoclonal antibody from cells generated in (a) hybridised with a fusion partner, e.g. non-secreting myeloma cells;
c. determining which hybridomas of (b) blocked IgA binding and taking these monoclonal antibodies.

3. A bifunctional antibody or heteroantibody comprising:

a. at least one antigen binding region e.g. a Fab fragment, derived from the human anti-Fc-alpha receptor antibody of claim 1; and
b. at least one antigen binding region e.g. a Fab fragment, specific for a target epitope.

4. The bifunctional antibody or heteroantibody according to claim 3 wherein

a. the target epitope is from a tumour cell; or
b. the target epitope is from a microorganism for example bacteria or a virus; or
c. the target epitope is from an auto-antibody producing lymphocyte or an IgE producing lymphocyte; or
d. the target epitope is on a soluble antigen, for example an auto-antibody or a rheumatoid factor.

5. The bifunctional antibody or heteroantibody according to claim 4, wherein the tumour cell is selected from myeloid leukaemia cells, small cell carcinoma of the lung cells, ovarian carcinoma cells and colon carcinoma cells.

6. A bifunctional antibody or heteroantibody according to any one of claims 3, 4, and 5, wherein the bifunctional antibody or heteroantibody is produced recombinantly.

7. A hybridoma cell line which produces the antibody of claim 1.

8. The monoclonal antibody of claim 1, or active fragment thereof, for use in therapy or diagnosis, for example: the treatment or diagnosis of autoimmune diseases, allergies or tumours.

9. Use of the monoclonal antibody of claim 1, or active fragment thereof, for the manufacture of a medicament for treatment or diagnosis of autoimmune diseases allergies or tumours.

10. The bifunctional antibody or heteroantibody of any one of claims 3 to 6 for use in therapy e.g. for use in tumour therapy; or for use in the treatment of autoimmune disease; or for use in the treatment of allergies.

11. Use of the bifunctional antibody or heteroantibody of any one of claims 3 to 6, for the manufacture of a medicament for the treatment of autoimmune disease or the treatment of allergies, or for the treatment of tumours.

Claims for the following Contracting State : ES

1. A method of producing a monoclonal antibody or an active fragment thereof, specific for human Fc receptor for IgA which antibody can trigger Fc-receptor-mediated effector cell activity comprising the sequential steps of:

a. immunizing an animal, e.g. a mouse with an Fc-alpha receptor e.g. human cells bearing the Fc-alpha receptor, lysates of said cells, or a purified preparation of the Fc-alpha receptor;
b. preparing hybridomas of anti monocyte monoclonal antibody from cells generated in (a) hybridised with a fusion partner, e.g. non-secreting myeloma cells;
c. determining which hybridomas of (b) specifically bind to the human Fc-alpha receptor and taking these monoclonal antibodies.

2. A method of producing a bifunctional antibody or heteroantibody comprising linking:

a. at least one antigen binding region e.g. a Fab fragment, derived from a monoclonal antibody specific for human Fc receptor for IgA which antibody can trigger Fc-receptor-mediated effective cell activity; with
b. at least one antigen binding region e.g. a Fab fragment, specific for a target epitope.

3. The method according to claim 2 wherein the bifunctional antibody or heteroantibody is produced recombinantly.

4. The method according to claim 2 wherein

   a. the target epitope is from a tumour cell; or
   b. the target epitope is from a microorganism for example bacteria or a virus; or
   c. the target epitope is from an auto-antibody producing lymphocyte or an IgE producing lymphocyte; or
   d. the target epitope is on a soluble antigen, for example an auto-antibody or a rheumatoid factor.

5. The method according to claim 4, wherein the tumour cell is selected from myeloid leukaemia cells, small cell carcinoma of the lung cells, ovarian carcinoma cells and colon carcinoma cells.

6. Use of a monoclonal antibody or active fragment thereof, specific for human Fc receptor for IgA which antibody can trigger human Fc-receptor-mediated effector cell activity, for the manufacture of a medicament.

7. Use of the bifunctional antibody or heteroantibody as defined in any one of claims 2 to 5, for the manufacture of a medicament.

8. Use according to claim 6 or claim 7 wherein the medicament is for the treatment or diagnosis of autoimmune diseases, allergies or tumours.

Patentansprüche

Patentansprüche für folgende Vertragsstaaten : AT, BE, CH, LI, DE, DK, FR, GB, IT, LU, NL, SE

1. Monoklonaler, für den menschlichen Fc-Rezeptor für IgA spezifischer Antikörper oder ein aktives Fragment davon, wobei der Antikörper die Fc-Rezeptor-vermittelte Effektorzellenaktivität auslösen kann.

2. Verfahren zur Herstellung des monoklonalen Antikörpers nach Anspruch 1 oder eines wirksamen Fragments davon, das die aufeinanderfolgenden Stufen umfaßt:

   a. Immunisieren eines Tieres, z.B. einer Maus, mit menschlichen, Fc-alpha-Rezeptor-tragenden Zellen;
   b. Herstellen von Hybridomen des monoklonalen Antimonozyten-Antikörpers aus in (a) erzeugten Zellen, die mit einem Fusionspartner, z.B. nicht sezernierenden Myelomzellen, hybridisiert sind;
   c. Bestimmen, welche Hybridome von (b) die IgA-Bindung blockierten und Heranziehen dieser monoklonalen Antikörper.

3. Bifunktioneller Antikörper oder Heteroantikörper, der umfaßt:

   a. wenigstens eine Antigen-Bindungsregion, z.B. ein Fab-Fragment, das aus dem menschlichen Anti-Fc-alpha-Rezeptor-Antikörper nach Anspruch 1 stammt; und
   b. wenigstens eine Antigen-Bindungsregion, z.B. ein Fab-Fragment, das für ein Ziel-Epitop spezifisch ist.

4. Bifunktioneller Antikörper oder Heteroantikörper, nach Anspruch 3, wobei

   a. das Ziel-Epitop aus einer Tumorzelle stammt; oder
   b. das Ziel-Epitop aus einem Mikroorganismus, beispielsweise aus Bakterien oder aus einem Virus, stammt; oder
   c. das Ziel-Epitop aus einem Autoantikörper-produzierenden Lymphozyten oder einem IgE-produzierenden Lymphozyten stammt; oder
   d. das Zielepitop sich auf einem löslichen Antigen, beispielsweise einem Autoantikörper oder einem Rheumafaktor, befindet.

5. Bifunktioneller Antikörper oder Heteroantikörper nach Anspruch 4, wobei die Tumorzelle ausgewählt ist aus myeloiden Leukämiezellen, Zellen von kleinzelligem Lungenzellen-Karzinom, Ovarialkarzionomzellen und Darmkarzinomzellen.

6. Bifunktioneller Antikörper oder Heteroantikörper nach einem der Ansprüche 3, 4 und 5, wobei der bifunktionelle

Antikörper oder der Heteroantikörper durch Rekombination erzeugt wird.

7. Hybridomzellinie, die den Antikörper nach Anspruch 1 produziert.

8. Monoklonaler Antikörper nach Anspruch 1 oder aktives Fragment davon zur Verwendung bei der Therapie oder Diagnose, beispielsweise bei der Behandlung oder Diagnose von Autoimmunerkrankungen, Allergien oder Tumoren.

9. Verwendung des monoklonalen Antikörpers nach Anspruch 1 oder des aktiven Fragments davon zur Herstellung eines Medikaments zur Behandlung oder Diagnose von Autoimmunerkrankungen, Allergien oder Tumoren.

10. Bifunktioneller Antikörper oder Heteroantikörper nach einem der Ansprüche 3 bis 6 zur therapeutischen Verwendung, z.B. zur Verwendung bei der Tumortherapie, oder zur Verwendung bei der Behandlung der Autoimmunerkrankung oder zur Verwendung bei der Behandlung von Allergien.

11. Verwendung des bifunktionellen Antikörpers oder Heteroantikörpers nach einem der Ansprüche 3 bis 6 zur Herstellung eines Medikaments zur Behandlung von Autoimmunerkrankung oder zur Behandlung von Allergien oder zur Behandlung von Tumoren.

Patentansprüche für folgenden Vertragsstaat : ES

1. Verfahren zur Herstellung eines monoklonalen, für den menschlichen Fc-Rezeptor für IgA spezifischer Antikörpers oder eines aktiven Fragments davon, wobei der Antikörper die Fc-Rezeptor-vermittelte Effektorzellenaktivität auslösen kann, das die folgenden aufeinanderfolgenden Stufen umfaßt:

   a. Immunisieren eines Tieres, z.B. einer Maus, mit einem Fc-alpha-Rezeptor, z.B. mit menschlichen Zellen, die den Fc-alpha-Rezeptor tragen, Lysaten dieser Zellen oder mit einer gereinigten Präparation des Fc-alpha-Rezeptors;
   b. Herstellen von Hybridomen des monoklonalen Antimonozyten-Antikörpers aus in (a) erzeugten Zellen, die mit einem Fusionspartner, z.B. nicht sezernierenden Myelomzellen, hybridisiert sind;
   c. Bestimmen, welche Hybridome von (b) spezifisch an den menschlichen Fc-alpha-Rezeptor binden und Heranziehen dieser monoklonalen Antikörper.

2. Verfahren zur Herstellung eines bifunktionellen Antikörpers oder eines Heteroantikörpers, das das Verknüpfen von folgendem umfaßt:

   a. wenigstens einer Antigen-Bindungsregion, z.B. einem Fab-Fragment, das von einem für den menschlichen Fc-Rezeptor für IgA spezifischen monoklonalen Antikörper stammt, wobei der Antikörper die Fc-Rezeptor-vermittelte Effektorzellenaktivität auslösen kann: mit
   b. wenigstens einer Antigen-Bindungsregion, z.B. einem für ein Ziel-Epitop spezifischen Fab-Fragment.

3. Verfahren nach Anspruch 2, wobei der bifunktionelle Antikörper oder Heteroantikörper durch Rekombination erzeugt wird.

4. Verfahren nach Anspruch 2, wobei

   a. das Ziel-Epitop aus einer Tumorzelle stammt; oder
   b. das Ziel-Epitop aus einem Mikroorganismus, beispielsweise aus Bakterien oder aus einem Virus, stammt; oder
   c. das Ziel-Epitop aus einem Autoantikörper-produzierenden Lymphozyten oder einem IgE-produzierenden Lymphozyten stammt; oder
   d. das Zielepitop sich auf einem löslichen Antigen, beispielsweise einem Autoantikörper oder einem Rheumafaktor, befindet.

5. Verfahren nach Anspruch 4, wobei die Tumorzelle ausgewählt ist aus myeloiden Leukämiezellen, Zellen von kleinzelligem Lungenzellen-Karzinom, Ovarialkarzionomzellen und Darmkarzinomzellen.

6. Verwendung eines monoklonalen, für den menschlichen Fc-Rezeptor für IgA spezifischen Antikörpers oder eines aktiven Fragments davon, wobei der Antikörper die durch den menschlichen Fc-Rezeptor vermittelte Effektorzellenaktivität auslösen kann, zur Herstellung eines Medikaments.

7. Verwendung des bifunktionellen Antikörpers oder des Heteroantikörpers nach einem der Ansprüche 2 bis 5 zur Herstellung eines Medikaments.

8. Verwendung nach Anspruch 6 oder Anspruch 7, wobei das Medikament zur Behandlung oder Diagnose von Autoimmunerkrankungen, Allergien oder Tumoren vorgesehen ist.

Revendications

Revendications pour les Etats contractants suivants : AT, BE, CH, LI, DE, DK, FR, GB, IT, LU, NL, SE

1. Anticorps monoclonal, ou fragment actif de celui-ci, spécifique du récepteur Fc humain d'une IgA, ledit anticorps pouvant déclencher une activité cellulaire effectrice médiée par le récepteur Fc.

2. Procédé de production de l'anticorps monoclonal selon la revendication 1 ou d'un fragment actif de celui-ci, comprenant les étapes séquentielles consistant à :

   a. immuniser un animal, par exemple une souris, avec des cellules humaines portant le récepteur Fc-alpha ;
   b. préparer des hybridomes d'anticorps monoclonal anti-monocyte à partir des cellules générées à l'étape (a), hybridées avec un partenaire de fusion, par exemple des cellules de myélome non sécrétrices ;
   c. déterminer quels hybridomes de l'étape (b) bloquent la liaison de l'IgA et prélever ces anticorps monoclonaux.

3. Anticorps bifonctionnel ou hétéroanticorps comprenant :

   a. au moins une région de liaison à un antigène par exemple un fragment Fab, dérivé de l'anticorps humain anti-récepteur-Fc-alpha de la revendication 1 ; et
   b. au moins une région de liaison à un antigène par exemple un fragment Fab, spécifique d'un épitope cible.

4. Anticorps bifonctionnel ou hétéroanticorps selon la revendication 3 dans lequel

   a. l'épitope cible est issu d'une cellule tumorale ; ou
   b. l'épitope cible est issu d'un microorganisme par exemple une bactérie ou un virus ; ou
   c. l'épitope cible est issu d'un lymphocyte producteur d'auto-anticorps ou d'un lymphocyte produisant des IgE ; ou
   d. l'épitope cible est présent sur un antigène soluble, par exemple un auto-anticorps ou un facteur rhumatoïde.

5. Anticorps bifonctionnel ou hétéroanticorps selon la revendication 4, la cellule tumorale étant choisie parmi les cellules de leucémie myéloïde, de cellules de carcinome de poumon à petites cellules, de cellules de carcinome ovarien et de cellules du carcinome du colon.

6. Anticorps bifonctionnel ou hétéroanticorps selon l'une quelconque des revendications 3, 4 et 5, l'anticorps bifonctionnel ou l'hétéroanticorps étant produit par voie recombinante.

7. Lignée cellulaire d'hybridome qui produit l'anticorps selon la revendication 1.

8. Anticorps monoclonal selon la revendication 1 ou fragment actif de celui-ci pour une utilisation en thérapie ou en diagnostic, par exemple : dans le traitement de diagnostic de maladies auto-immunes, allergies ou tumeurs.

9. Utilisation de l'anticorps monoclonal selon la revendication 1 ou fragment actif de celui-ci, pour la fabrication d'un médicament pour le traitement ou le diagnostic de maladies auto-immunes, d'allergies ou de tumeurs.

10. Anticorps bifonctionnel ou hétéroanticorps selon l'une quelconque des revendications 3 à 6 pour une utilisation

en thérapie, par exemple pour une utilisation en thérapie antitumorale ; ou pour une utilisation dans le traitement de maladie auto-immune ; ou pour l'utilisation dans le traitement des allergies.

11. Utilisation de l'anticorps bifonctionnel ou hétéroanticorps selon l'une quelconque des revendications 3 à 6 pour la fabrication d'un médicament pour le traitement de maladie auto-immune ou le traitement des allergies ou pour le traitement des tumeurs.

Revendications pour l'Etat contractant suivant : ES

1. Procédé de production d'un anticorps monoclonal ou d'un fragment actif de celui-ci, spécifique d'un récepteur Fc humain pour une IgA, ledit anticorps pouvant déclencher une activité cellulaire effectrice médiée par un récepteur Fc, ledit procédé comprenant les étapes séquentielles consistant à :

   a. immuniser un animal, par exemple une souris avec un récepteur Fc-alpha, par exemple avec des cellules humaines portant le récepteur Fc-alpha, les lysats desdites cellules, ou une préparation purifiée du récepteur Fc-alpha ;
   b. préparer des hybridomes d'anticorps monoclonal anti-monocyte à partir des cellules générées à l'étape (a), hybridées avec un partenaire de fusion, par exemple des cellules de myélome non sécrétrices ;
   c. déterminer quels hybridomes de l'étape (b) se lient spécifiquement au récepteur Fc-alpha humain et prélever ces anticorps monoclonaux.

2. Procédé de production d'un anticorps bifonctionnel ou d'un hétéroanticorps comprenant la réticulation :

   a. d'au moins une région de liaison à un antigène par exemple un fragment Fab, dérivé de l'anticorps spécifique d'un récepteur Fc humain pour une IgA, ledit anticorps pouvant déclencher une activité cellulaire effectrice médiée par un récepteur Fc ; avec
   b. au moins une région de liaison à un antigène par exemple un fragment Fab, spécifique d'un épitope cible.

3. Procédé selon la revendication 2 dans lequel l'anticorps bifonctionnel ou l'hétéroanticorps est produit par voie recombinante.

4. Procédé selon la revendication 2 dans lequel

   a. l'épitope cible est issu d'une cellule tumorale ; ou
   b. l'épitope cible est issu d'un microorganisme par exemple une bactérie ou un virus ; ou
   c. l'épitope cible est issu d'un lymphocyte producteur d'auto-anticorps ou d'un lymphocyte produisant des IgE ; ou
   d. l'épitope cible est présent sur un antigène soluble, par exemple un auto-anticorps ou un facteur rhumatoïde.

5. Procédé selon la revendication 4, dans lequel la cellule tumorale est choisie parmi les cellules de leucémie myéloïde, de cellules de carcinome de poumon à petites cellules, de cellules de carcinome ovarien et de cellules du carcinome du colon.

6. Utilisation d'un anticorps monoclonal ou d'un fragment actif de celui-ci, spécifique d'un récepteur Fc humain pour une IgA, ledit anticorps pouvant déclencher une activité cellulaire réceptrice médiée par un récepteur Fc, pour la préparation d'un médicament.

7. Utilisation d'un anticorps bifonctionnel ou hétéroanticorps selon l'une quelconque des revendications 2 à 5, pour la fabrication de médicament.

8. Utilisation selon la revendication 6 ou la revendication 7 dans laquelle le médicament est destiné au traitement et au diagnostic de maladies auto-immunes, d'allergies ou de tumeurs.